Heat-Shock Transformation

Materials and Equipment

- Drigalski spatula or glass beads
- Alcohol lamp
- Microtube rack
- Lighter
- Micropipette
- Micropipette tips
- Microtubes (0.6 ml)
- Shaker
- Incubator
- Laminar flow hood
- Thermoblock or water bath

Reagents

- Plasmid DNA
- Chemical competent cells
- SOC medium
- Plaques with LB agar and proper antibiotic (KAN 15 mg/ml, CAM 35 mg/ml, AMP 100 mg/ml)
- Ethanol 96%
- Liquid LB medium

Methodology

1. As a previous step, the thermoblock (or water bath) at 42˚C has to be programmed.
2. Take an aliquot of chemical competent cells and defrost in ice.
3. Add 5µl of pDNA (concentration between 200-300 pg/ml) to 50 µl of competent cells.
4. Incubate on ice for 30 minutes.
5. Heat shock (thermoblock or water bath) at 42˚C for 30 seconds.
6. Place samples on ice for 5 minutes.
7. Add 950µl of SOC medium in sterile conditions.
8. Incubate at 37˚C for 1 hour, 250 rpm.
9. Plate 200 µl of transformed cells into warm LB agar plates with the proper antibiotic.
10. Incubate ON at 37˚C. (12h-16h)
11. Isolate a single colony and culture in liquid LB broth for future procedures.